Pharmacology Biochemistry and Behavior, Vol. 60, No. 2, pp. 415–421, 1998

0091-3057/98 $19.00 + .00

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PII S0091-3057(97)00599-6

Training Dose and Session Time as Contextual Determinants of Heroin Self-Administration in Rats

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Received 28 March 1997; Revised 30 October 1997; Accepted 12 November 1997

MARTIN, T. J., J. E. SMITH AND S. I. DWORKIN. *Training dose and session time as contextual determinants of heroin self-administration in rats*. PHARMACOL BIOCHEM BEHAV **60**(2) 415–421, 1998.—In this study a rat self-administration model was used to examine the effects of training dose and time in the session on the dose–effect curve for heroin. Doses of heroin lower than 5.4 μg/inf maintained higher rates of drug intake in animals trained with 5.4 μg/inf compared to 18 μg/inf. Doses greater than 5.4 μg/inf maintained similar rates of intake in both groups of animals. The dose–response curve was shifted downward and to the right as the session progressed for animals trained with 5.4 μg/inf of heroin; however, the shift in the dose–intake curve over the session was less pronounced when the training dose was 18 μg/inf. Naltrexone and naltrindole were administered to animals in which responding was engendered with infusions of 5.4 μg of heroin to determine the effects of these antagonists in the context of time is the session. The potency of naltrexone decreased across the 4 h of the session with a time course that was consistent with literature reports on the elimination kinetics of naltrexone in rat brain. In contrast, there was not a significant interaction between naltrindole dose and session time. Therefore, the rates of heroin intake in rats are dependent not only upon the dose available for self-administration, but upon the session time and training dose as well.

Reinforcement Antagonism Naltrexone Naltrindole Opioid μ-Opioid δ-Opioid

NONHUMAN drug self-administration studies allow for a valid assessment of compulsive drug use in humans. Data from these studies have provided significant information on variables that can effect drug intake. Both the pharmacological effects of the drug and factors related to drug availability can modulate the behavioral and neurochemical effects of a substance (10). Thus, the rate of responding maintained by drugs results from a complex interaction of pharmacological actions and behavioral effects. One complicating factor in the evaluation drug self-administration experiments is the complex nature of the dose–effect curves. Low unit doses of a drug reinforcer will maintain little or no responding. As the dose is increased beyond some threshold, the drug will maintain high rates of responding. Increasing the dose further results in decreases in response rates due to pharmacological effects that interfere with responding, such as locomotor effects, as well as an increase in the duration of action of the larger doses compared to smaller doses. This complex dose–effect relationship arises due the interaction of effects of the drug that increase responding (e.g., reinforcing effects, stimulant effects) and effects that decrease responding (e.g., stereotypic locomotor effects, catalepsy). Therefore, drug self-administration experiments generate complex dose–effect data as a result of the interaction of various pharmacological effects (10).

Drug infusions can become associated with stimuli other than the pharmacological effects of the drug, and the interaction of these stimuli with drug effects may alter the rate of drug intake. These effects can, therefore, be associated with the context of drug administration. One contextual determinant of response rate-altering effects of a drug appears to be the time within the session during which responding is maintained by the drug (3). Although drug reinforcers can maintain relatively constant rates of responding during experimental sessions (8,9,12), response rates have been shown to vary

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across the session for the self-administration of some drug reinforcers under certain conditions. For example, a decrease in responding occurs over the course of self-administration sessions with either cocaine (7,9) or codeine (3,7). Furthermore, the decline in responding maintained by codeine as the self-administration session progresses does not occur when responding is maintained by food during components that are interspersed with components of codeine availability, demonstrating the modulatory role of the context of drug administration (3). These effects may be due to acute tolerance to the reinforcing effects or to an increase in the rate-decreasing effects of the drug during the session.

Although opiates have been studied as reinforcers in laboratory animals, contextual determinants of the interaction between session time and drug dose has not been documented. Heroin will maintain responding in rodents, an effect that is mediated by central μ- and perhaps δ-opioid receptors (12, 16). The patterns of responding maintained by heroin are dependent upon the dose available for self-administration. For example, 60 μg/kg of heroin will maintain relatively constant rates of responding during 3-h sessions, whereas smaller doses produce patterns characterized by multiple drug infusions being administered in relatively short periods of time followed by longer pauses (8,16). Although the delivery of infusions is dispersed throughout the session for a wide range of doses of heroin, the density of infusion delivery appears to change as the session progresses (8,16). The effects of antagonists on heroin self-administration also appear to change over the course of the session based upon published patterns of responding; however, rates of responding have not been analyzed with respect to time in the session (8,16). It may be difficult to evaluate the effects of antagonist treatment over session time without understanding how the dose–response function for heroin self-administration is altered with respect to session time. Although averaging the number of infusions over entire 3- to 4-h sessions may provide a reasonable approximation of overall antagonist effect, should the interaction of session time be dependent upon individual agonist dose, effects of antagonists may be obscured by such approximations.

These experiments were initiated to determine if the time within the session is a contextual determinant of rates of heroin self-administration and the extent to which the evaluation of antagonist effects differs when time in the session is taken into account. Infusions of 5.4 μg of heroin were used to engender and maintain responding in rats and the dose–response relationship for heroin was evaluated by substituting seven doses (0.3 to 30 μg/inf) during test sessions. The relationship between number of infusions administered and heroin dose was analyzed for each of the four hours of the session. These data were compared to data obtained from animals in which responding was engendered and maintained by infusions of 18 μg to determine if the training dose altered this contextual relationship between session time and the heroin dose–response relationship. The 5.4 μg dose of heroin was chosen because it is at the apex of the dose–effect curve and maintains a pattern of responding that is characterized by multiple infusions being taken within short periods of time followed by long pauses (8). The 18 μg/infusion dose was chosen because it maintains a pattern of responding that is characterized by regular interinfusion intervals that is qualitatively dissimilar to that maintained by 5.4 μg/infusion (14). The 18 μg/infusion dose was also chosen because it has been frequently used in the literature (60 μg/kg/infusion) to characterize heroin self-administration in rats (8,16). The effects of naltrexone and naltrindole were also evaluated on responding maintained by infusions of 5.4 μg of heroin to determine if session time was a contextual determinant of their alteration of heroin self-administration. These antagonists attenuate the reinforcing effects of 60 μg/kg of heroin in rats (8,12,16), but their effects with respect to time in the self-administration session have not been evaluated. These data will hopefully determine the extent to which the reinforcing properties of heroin as well as the effects of naltrexone and naltrindole are dependent upon the time within a session as well as the training dose of heroin. Such variables may, therefore, become important considerations in the design and analysis of heroin self-administration experiments.

METHODS

Subjects

Sixteen Male, Fischer 344 rats (approx. 90 days old at the beginning of the experiments, 250 to 300 g) were used for these experiments and kept at 85% of their free-feeding body weight to maintain health and reduce weight fluctuations (1). Food restriction also increases drug acquisition and maintenance of stable patterns of responding (4–6). The animals were placed on a reversed light–dark cycle (dark 0500–1700), and all behavioral sessions were conducted during the dark phase of the cycle. Animals were housed in acrylic cages (24 × 26 × 21 cm) in ventilated sound-attenuated chambers with ad lib access to water except during experimental sessions. Each animal was implanted with an external jugular vein catheter as described previously (14,17) under anesthesia induced with atropine (10 mg/kg, IP) and pentobarbital (50 mg/kg, IP). Animals were administered penicillin G procaine (75,000 U, IM), and exterior wounds were dressed with antibiotic powder. Catheter patency was maintained by hourly infusions of saline with heparin (1.7 U/ml) and checked periodically with methohexital (10 mg/kg). If patent, loss of consciousness occurred within 5 s of methohexital infusion.

Apparatus

All experimental sessions were conducted in sound-attenuated chambers and were controlled by an IBM-compatible computer through an interface (MED Associates, St. Albans, VT). The acrylic housing cage served as the experimental chamber for all animals. The animal was transported in the housing cage to a sound-attenuated enclosure where a lever was inserted into the chamber and a light above the lever was connected to the computer interface. Each sound-attenuated enclosure was equipped with a house light, tone generator, ventilator fan, and infusion pump (Razel Inc., Stamford, CT). The fluid swivel and catheter were flushed with 0.5 ml of heparinized saline and connected to the infusion pump.

Behavioral Training

Responding was engendered in rats with infusions of either 5.4 ($n = 8$) or 18 μg ($n = 8$) of heroin in a volume of 0.2 ml under an FR 1 schedule of reinforcement. Illumination of the light above the lever indicated drug availability and a 30-s time out followed each infusion, during which the lever light was darkened and the house light and tone generator were operated. Responses during the last 5 s of the time-out period reset the time out by 5 s. Once stable responding was obtained, defined as five successive sessions during which the number of infusions did not vary by more than 10% of the mean, the FR value was gradually increased over several ses-

sions to a terminal value of 10. Sessions were 4 h in duration, and were conducted on Monday through Friday.

Dose–Effect Determinations for Heroin

After a minimum of 5 days of stable responding under the FR 10 schedule, dose-intake manipulations were initiated. Several doses of heroin (0.3, 0.9, 1.8, 5.4, 9, 18, or 30 μg/inf) or saline were substituted in random order for the training dose on Tuesdays or Thursdays if the number of infusions administered during the previous session did not vary by more than 10% of the mean for the training dose. Following substitution of all doses, a duplicate determination of the heroin dose–effect curve was performed in all animals in a similar manner.

Effect of Naltrexone and Naltrindole on Heroin Self-Administration

The effects of naltrexone or naltrindole were evaluated over the four hours of the session in the eight animals trained to self-administer 5.4 μg/inf of heroin. Animals were pretreated with vehicle (0.3% ethanol), naltrexone (0.03, 0.1, 1, or 10 mg/kg) or naltrindole (1, 3, 10, or 30 mg/kg) SC 10 min prior to the session on Tuesdays or Thursdays provided that the number of infusions of heroin did not vary by more than 10% from the mean for the training dose on the previous day. Vehicle was tested first in all animals. The order of antagonist doses was randomized, and the order of antagonist tested was counterbalanced such that naltrexone was tested prior to naltrindole in half of the animals. Double determinations were made for each dose of antagonist and vehicle.

Statistics

The data for each hour of the session were evaluated using a two-way ANOVA with heroin dose and hour being the independent variables and number of infusions being the dependent measure. The influence of the training dose on the heroin dose–response curves was evaluated for the entire session using a two-way ANOVA, with heroin dose and training dose being the independent measures and number of infusions being the dependent variable. The data from the antagonist experiments were evaluated similarly using a two-way ANOVA, with antagonist dose and session hour being the independent variables and number of infusions being the dependent measure. Post hoc comparisons were made using Fischer's protected least significant differences for multiple comparisons. The α level for all statistical analyses was 0.05.

Drugs and Chemicals

Heroin hydrochloride was provided by the National Institute on Drug Abuse and dissolved in 0.9% (w/v) saline with heparin (1.7 U/ml). Atropine sulfate was purchased from Sigma Chemical Co. (St. Louis, MO) and dissolved in water. Naltrindole was a gift from Burroughs-Wellcome Inc. (Research Triangle Park, NC) and was dissolved in 0.3% ethanol. Naltrexone hydrochloride was purchased from Sigma Chemical Co., and was dissolved in 0.3% ethanol. Pentobarbital (Nembutal®) was purchased from Abbott Laboratories (North Chicago, IL) in a vehicle of 10:40:50 (v/v) ethanol:propylene glycol:water. Methohexital (Brevital®) was purchased from Eli Lilly Co. (Indianapolis, IN) and dissolved in sterile water. Antibiotic powder (Polysporin®) was purchased from Burroughs-Wellcome Inc. and heparin was purchased from Elkins-Sinn Co. (Cherry Hill, NJ).

RESULTS

Dose–Response Curves for Heroin Self-Administration

Responding was maintained in a dose-responsive manner in animals trained to self-administer infusions of either 5.4 or 18 μg of heroin. The dose–intake relationship, however, was significantly different between the groups that were trained on 5.4 μ/infusion compared to those trained with 18 μg/infusion. The number of infusions administered was a biphasic function of dose when responding was engendered by 5.4 μg/inf of heroin, $F(7, 48) = 18.68$, $p \leq 0.0001$ (Fig. 1). The number of infusions administered was greater for all doses of heroin in this group of animals compared to the number of infusions of saline ($p \leq 0.05$). The number of infusions administered was also a biphasic function of dose when responding was engendered with infusions of 18 μg/inf, $F(7,32) = 26.08$, $p \leq .0001$. However, self-administration of the two lowest doses of heroin (0.3 and 0.9 μg/inf) was not significantly greater than saline for these animals ($p \leq 0.05$). The training dose significantly altered the dose–response curve for the number of infusions administered across the range of heroin doses studied, $F(1,80) = 22.929$, $p = 0.0001$. The three lowest doses of heroin (0.3, 0.9, and 1.8 μg/inf) and the 9 μg dose maintained higher response rates ($p \leq 0.05$) when responding was engendered with infusions of 5.4 μg compared to when responding was engendered with infusions of 18 μg of heroin.

Hourly Assessment of Dose–Response Curves for Heroin Self-Administration

The dose–response relationship for the number of infusions administered decreased across the session when responding was engendered with infusions of 5.4 μg of heroin, $F(21, 187) = 3.174$, $p \leq 0.0001$ (Fig. 2). The number of infusions administered in the first session hour was dose dependent, $F(7, 46) = 9.661$, $p \leq 0.0001$, and all doses maintained rates of responding significantly greater than saline ($p \leq 0.05$). The number of infusions administered was lower in the second session hour compared to the first ($p = 0.0001$), but was also dependent on the heroin dose, $F(7, 46) = 3.624$, $p =$

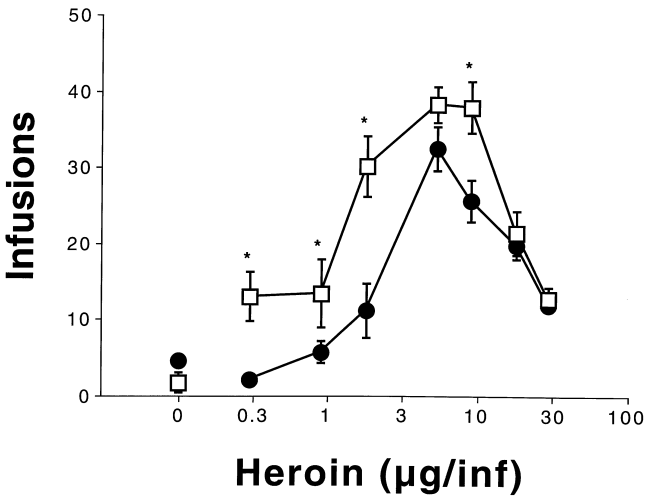

FIG. 1. Dose–response relationship for heroin self-administration. The mean (SEM) number of infusions administered over the entire session are shown for animals trained with infusions of 5.4 (□, $n = 7$) or 18 (●, $n = 5$) μg of heroin. *Significantly different for the two training doses, $p \leq 0.05$.

0.0033. All doses maintained response rates that were greater than that maintained by saline in the second hour ($p \leq 0.05$). The number of infusions administered in the third hour of the session was dose dependent, $F(7, 46) = 7.095$, $p \leq 0.0001$, but decreased compared to both the first ($p = 0.0001$) and second ($p = 0.028$) hour. In the third hour of the session, however, doses lower than 5.4 μg/inf of heroin did not maintain rates of responding that were greater than that maintained by saline, and only a descending limb of the dose–response curve was apparent. The number of infusions administered in the fourth session hour was dose dependent, $F(7, 46) = 5.192$, $p = 0.0002$, and further decreased compared to the first ($p = 0.0001$), second ($p = 0.0001$), and third ($p = 0.029$) hours. As with the third session hour, doses lower than 5.4 μg/inf of heroin did not maintain responding at a rate greater than that obtained with saline. Therefore, the ascending limb of the dose–response curve for heroin self-administration is shifted downward and to the right across the 4 h of the session when infusions of 5.4 μg are used to engender responding, whereas the descending limb is relatively unchanged until the fourth session hour.

In contrast to the above findings, the dose–response curve for the number of infusions administered was decreased only in the fourth hour of the session when responding was engendered with infusions of 18 μg of heroin (Fig. 3). As with the data obtained from animals in which responding was engendered with 5.4 μg/inf of heroin, the dose–response curve for the number of infusions administered was dependent upon the session hour when responding was engendered with infusions of 18 μg of heroin, $F(21, 129) = 1.985$, $p = 0.011$. However, the number of infusions administered was not different in the second and third hours compared to the first. The number of infusions administered in the fourth session hour was less compared to the first ($p = 0.0001$), second ($p = 0.0083$), and third ($p = 0.0077$) hour. The number of infusions administered in the first session hour was dependent upon the heroin dose, $F(7, 33) = 4.823$, $p = 0.0008$; however, 0.3 μg/inf heroin did not maintain responding at a rate greater than that maintained by saline. The number of infusions administered in the second session hour was dose dependent $F(7, 33) = 19.11$, $p \leq 0.0001$, and also a biphasic function of dose, because 1.8 μg/inf of heroin maintained a rate of responding that was greater than that maintained by saline but less than that maintained by 5.4 μg/inf. Doses lower than 1.8 μg/inf did not maintain rates of responding greater than that maintained by saline in the second session hour. Heroin maintained responding in the third session hour in a dose-dependent manner as well, $F(7, 33) = 18.05$, $p \leq 0.0001$. As with responding engendered by 5.4 μg/inf of heroin, the number of infusions administered was a decreasing function of these doses of heroin in the third session hour because doses lower than 5.4 μg/inf did not maintain response rates greater than those maintained by saline. The number of infusions administered in the fourth session hour was dose dependent $F(7, 33) = 14.481$, $p \leq 0.0001$, but was significantly less than the first $p \leq 0.0001$), second ($p = 0.0077$), and third ($p = 0.0083$) session hours. As in the third session hour, doses lower than 5.4 μg/inf did not maintain responding at rates significantly greater than that maintained by saline. Therefore, doses on the ascending limb of the dose–response curve for heroin maintained lower rates of responding when lever presses were engendered with infusions of 18 μg compared to 5.4 μg of heroin. Furthermore, the ascending limb of the dose–response curve was shifted downward only in the fourth session hour when infusions of 18 μg of heroin were used to engender responding.

Effects of Naltrexone and Naltrindole

Both naltrexone and naltrindole altered responding maintained by infusion of 5.4 μg of heroin. Both antagonists decreased the number of infusions delivered over the entire session, and naltrexone appeared to be slightly more potent than naltrindole. The effects of each antagonist changed over the 4 h of the session, however. The interaction between dose and session hour was significant for naltrexone, $F(12, 46) = 23.11$, $p = 0.021$, and the dose–response curve for naltrexone was shifted to the right with time (Fig. 4). Furthermore, the curve is shifted downward when the number of infusions is the de-

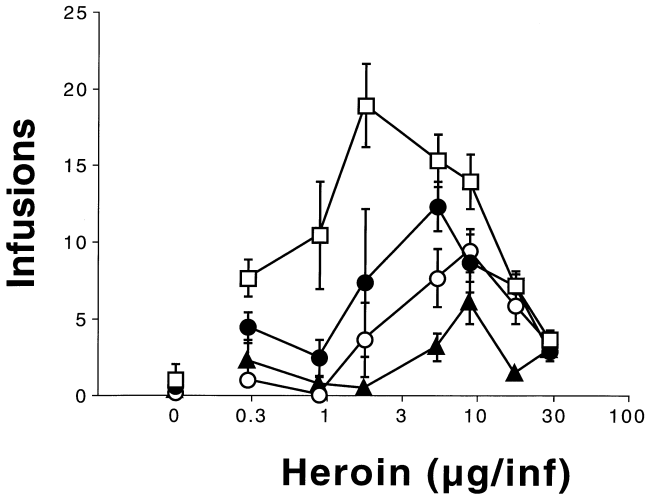

FIG. 2. Dose–response relationship for animals trained with 5.4 μg/inf of heroin for each session hour. The mean (SEM) number of infusions administered during the first (□), second (●), third (○), or fourth (▲) session hour are shown. The dose–response curves were significantly different for each session hour (see Results for statistics).

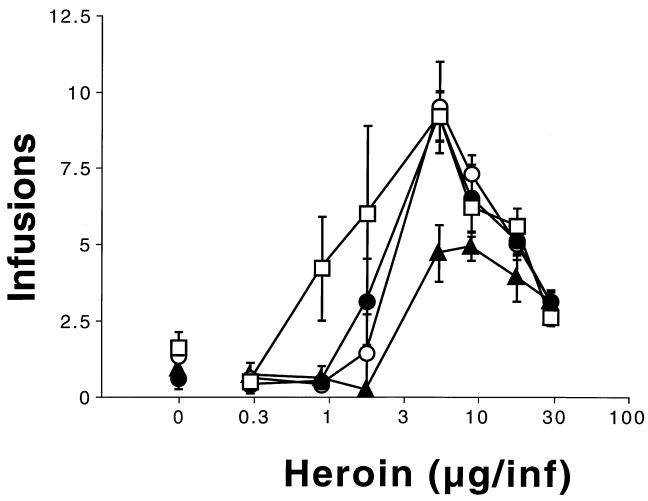

FIG. 3. Dose–response relationship for animals trained with 18.0 μg/inf of heroin for each session hour. The mean (SEM) number of infusions administered during the first (□), second (●), third (○), or fourth (▲) session hour are shown. The dose–response curve was significantly decreased in the fourth hour (see Results for statistics).

pendent measure, but not when the number of infusions are represented as the percentage of vehicle control, because the number of infusions administered under both control and vehicle conditions decrease with session time. In the first session hour, naltrexone altered the number of infusions administered in a dose-responsive manner, $F(4, 15) = 19.059$, $p \leq 0.0001$, with the number of infusions being significantly increased following pretreatment with 0.03 mg/kg and significantly decreased at the higher doses compared to vehicle. Naltrexone likewise had significant effects in the second session hour that were dose dependent, $F(4, 15) = 3.400$, $p = 0.036$. All doses other than 0.1 mg/kg decreased the number of infusions administered in the second hour compared to vehicle treatment. By the third hour of the session, only the highest dose of naltrexone significantly altered the number of infusions administered. In the fourth session hour, 1.0 mg/kg of naltrexone significantly increased responding, whereas 10 mg/kg decreased the number of infusions delivered in this hour of the session. The effects of naltrindole also changed over the course of the session, $F(5, 85) = 5.838$, $p \leq 0.0001$, but the interaction between session hour and dose was not significant, $F(13, 85) = 1.541$, $p = 0.12$, as all doses were similarly affected by session time (Fig. 5). None of the doses of naltrindole increased responding in any session hour, and only the highest dose (30.0 mg/kg) significantly decreased responding maintained by 5.4 μg/inf of heroin.

DISCUSSION

The results of this study indicate that training dose and time in the session are contextual variables that influence the self-administration of heroin. The dose–effect relationship for responding maintained by heroin was shifted downward and to the right over the course of a 4-h session, and the degree of this shift was dependent upon the dose of heroin used to maintain self-administration. In addition, there was a significant interaction between the effects of session time and the effects of naltrexone but not naltrindole. These data indicate that it may be useful to consider time in the session when evaluating the effects of antagonists on doses of heroin that are near the apex of the dose–effect curve.

Understanding the reasons for the shift in the dose–effect curves for heroin across the session is complicated by the fact that numerous factors influence the shape of dose–effect curves in drug self-administration experiments, and the interactions of these variables are poorly understood. A detailed analysis of the literature regarding these points is beyond the scope of this discussion; however, an excellent review has been provided elsewhere (11). Dose–effect curves for drug reinforcers are influenced not only by their rewarding effects, but also by other pharmacological effects such as locomotor effects, catalepsy or production, of aversive effects such as respiratory depression with opioids (11). These effects, in part, contribute to the decreases in responding that occur with

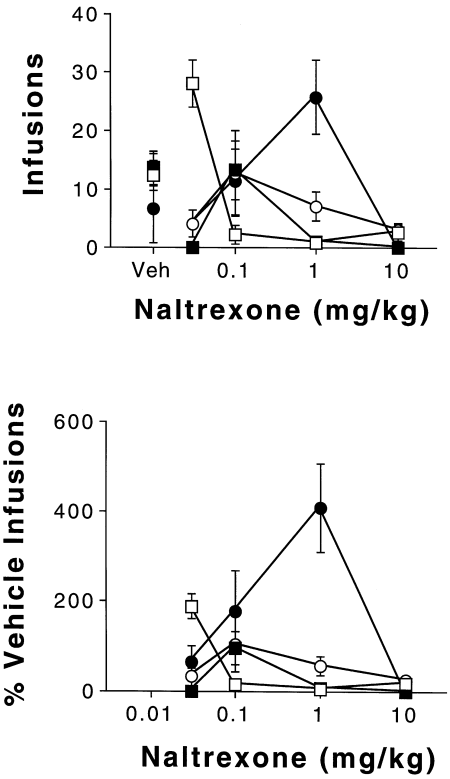

FIG. 4. Effects of naltrexone on responding maintained by 5.4 μg/inf of heroin during each session hour. The mean (SEM) number of infusions administered during the first (□), second (●), third (○), or fourth (■) session hour are shown (upper graph). The lower graph expresses the data as % vehicle-injected control for the individual session hours. Vehicle injections had no effect on the number of infusions taken in any hour and the potency of naltrexone decreased significantly during the session (see Results for statistics) $n = 8$.

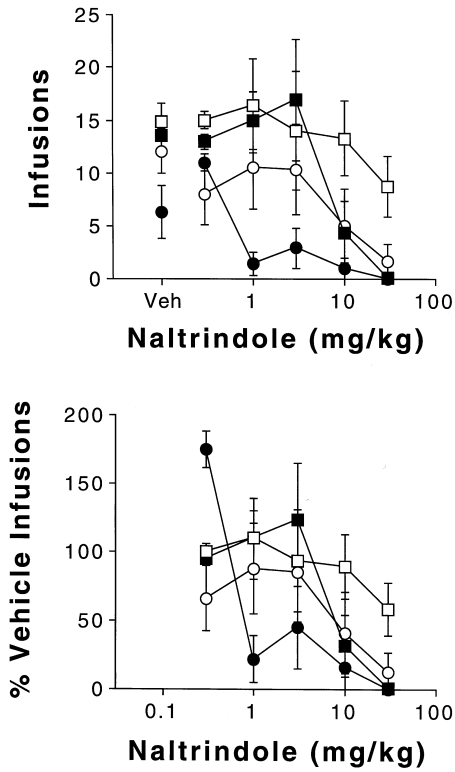

FIG. 5. Effects of naltrindole on responding maintained by 5.4 μg/inf of heroin during each session hour. The mean (SEM) number of infusions administered during the first (□), second (●), third (○), or fourth (■) session hour are shown (upper graph). The lower graph expresses the data as % vehicle-injected control for the individual session hours. Vehicle injections had no effect on the number of infusions taken in any hour (see Results for statistics) $n = 8$.

higher doses of drugs in self-administration experiments, and hence, the descending limb of the dose–effect curve. Pharmacokinetics and drug satiety have also been implicated in the descending limb of the dose–effect curve for drugs (10). Other factors have been found to affect the dose–response curves for drug reinforcers in self-administration paradigms. For example, the duration of time-out following infusions has been shown to influence the dose–response curve for responding maintained by cocaine (18). The same range of cocaine doses can produce either ascending, descending, or biphasic dose–response curves when the time-out values are large, small, or intermediate, respectively. Therefore, parameters relating to the schedule of reinforcement can influence the relationship between drug dose and response rate in self-administration paradigms. The changes that occur in the number of infusions administered with respect to time in the session may involve all or some of these pharmacological or behavioral effects.

Decreased drug intake across a self-administration session has been suggested to occur as a result of the rate-decreasing effects of drugs that increase with session time or as a result of drug satiety (11). This does not seem to be an adequate explanation for the present data, because the greatest decrease in response rates occur with lower doses of heroin. In fact, a decrease in response rates occur with lower doses of heroin. In fact, a decrease in the number of infusions delivered with respect to time within the session did not result when the highest dose of heroin was available. The present data suggest that the decrease in responding across the session is due to a loss in the reinforcing efficacy of the lower doses of heroin as the session progresses, because the dose–effect curves are shifted downward and to the right, and this shift occurs to a greater extent on the ascending compared to the descending limb of the dose–effect curve. Acute tolerance to the reinforcing effects of heroin does not appear to adequately address the current data because the decreases in number of infusions was less as the dose of heroin was increased. The rate of the development of tolerance is usually dose dependent, with higher doses producing tolerance at a greater rate and to a greater extent than lower doses. Therefore, it seems likely that the phenomenon of decreased number of infusions with time in the session is due to both pharmacological and behavioral effects of these doses of heroin.

The training dose modified the extent to which response rates decreased across the session, indicating that drug history may have some role in the patterns of responding that are maintained by heroin in rats. If the effect of session time on the dose–effect relationship for self-administration was due only to an alteration in pharmacological variables, one would not expect the training dose to alter this effect. However, the higher training dose clearly resulted in less of a decrease in responding across the session for the doses of heroin that were on the descending limb of the dose–effect curve. Furthermore, it also affected the doses on the ascending limb in that the doses lower than 5.4 μg/inf did not maintain responding after the initial hour of the session and the threshold dose of heroin that would maintain responding above that maintained by saline was higher. This may be due to the ability of the animals trained on the lower heroin dose to discriminate lower doses of heroin compared to the animals trained on 18 μg/infusion; however, such an hypothesis would need to be addressed with further studies. Regardless of the reason, the present data indicate that the ability of a particular dose of heroin to maintain responding as well as the pattern of responding that is maintained depends upon the training dose as well as the time in the session.

The effect of time in the session on the dose–effect curve for heroin self-administration differs between the present study and earlier work from our laboratory using a schedule of reinforcement than determines dose–effect curves in a single session (14). In this earlier manuscript, doses of heroin on the descending limb of the curve were made available for self-administration for 1 h each in increasing, decreasing or random orders of presentation with each dose occurring only once. When the lowest dose available, 5.4 μg/infusion (18 μg/kg/infusion), occurred in the third or fourth session hour the number of infusions administered increased compared to the first or second hour (14). Because these doses were available only once, however, this dose had been preceded by higher doses. Therefore, training under a different schedule of reinforcement than used for the present study produces a different effect of time in the session on number of infusions delivered of these doses of heroin on at least the descending limb of the dose–effect curve. Again, these effects are difficult to explain based upon the pharmacology of heroin only, and seem to indicate reinforcement schedule and training conditions as salient influences on the number of infusions administered with respect to time in the session for heroin.

The examination of the effects of naltrexone with respect to time in the session indicated that the potency of naltrexone decreased across the 4 h of the session. These findings are consistent with the time course of the biodisposition of naltrexone in rat brain following systemic injection (15). These investigators demonstrated that peak brain levels of [15,16-^{3}H]naltrexone occur 30 min following SC injection in rats, and that these levels decreased rapidly in the first 2 h following injection. Therefore, one would expect that the dose–effect curve for naltrexone would be shifted to the right during the course of a 4-h session. This is clearly shown by the data in Fig. 4. The dose–effect curve for naltrexone appears to be shifted downward and to the right as the session progresses when infusions are plotted as the dependent variable (upper graph in Fig. 4). The dose–effect curve is shifted upward and to the right when the negative acceleration with session time is factored into the analysis of the data by plotting the data as a percentage of vehicle-injected controls (lower graph in Fig. 4). The upward shift results from an increase in responding that occurred during the session following pretreatment with both 0.1 and 1.0 mg/kg of naltrexone, whereas a decrease occurred under both control conditions and following vehicle administration. This increase could result from a combination of antagonism of the reinforcing effects of heroin in the early parts of the session and antagonism of the rate-decreasing effects in the later session hours. It is noteworthy that the number of infusions administered in the first session hour following pretreatment with 1.0 mg/kg of naltrexone (25.7 ± 8.7) was similar to the number of infusions administered in the first session hour of control sessions (15.3 ± 1.7) or following vehicle injection (14.9 ± 1.6). The decrease in the effects of naltrexone over the course of the session may also be due to increased brain levels of heroin and its metabolites during the session, which would more effectively compete for binding to the receptor sites. These data, therefore, indicate that time in the session is also a variable that may be considered when evaluating the effects of naltrexone on heroin self-administration.

The effects of naltrindole were not altered over the course of the session. Responding maintained by 5.4 μg/inf of heroin decreased over all hours of the session following pretreatment with all doses of naltrindole. Only decreases in responding were observed and only at relatively high doses (10 and 30 mg/kg) of naltrindole. This range of doses is consistent with those

that increase responding maintained by 60 μg/kg/inf of heroin in rats (16). Time course studies with naltrindole suggest that its effects are of short duration, reaching peak effects in 40 min following SC injection (2). Therefore, the lack of interaction with session time likely results from effects in the first hour only. A decrease in responding occurred following administration of all doses of naltrindole that was comparable to that observed in control sessions and following vehicle injection. It may be that no increases in the rate of responding maintained by 5.4 μg/inf of heroin occurred following naltrindole administration because δ-opioid receptors are less involved in the rate-decreasing effects of this dose of heroin. A decrease in responding would be consistent with an attenuation of the reinforcing effects of this dose of heroin, because it is at the apex of the dose–effect curve. Therefore, these data seem consistent with the literature reports that δ-opioid receptors may be involved in heroin reinforcement. Clearly it would be advantageous to investigate the involvement of δ-opioid receptors in heroin reinforcement further using more potent compounds with longer duration of action that have been recently developed (13) or under different schedules of heroin reinforcement.

In conclusion, the dose–response relationship for the rate of responding and the rate of reinforcement maintained by heroin is dependent upon session time, and this relationship is dependent upon the dose of heroin used to engender responding. The antagonism data suggest that opioid receptors may be involved in the behavioral effects that are altered by the contextual variables examined in this study.

ACKNOWLEDGEMENTS

This research was supported by the National Institute on Drug Abuse of the National Institutes of Health through grants DA-00247 (T.J.M.), DA-01999 (J.E.S.), DA-06634 (J.E.S.), and DA-00114 (J.E.S.). The work reported in this manuscript was conducted in accordance with the Guide for the Care and Use of Laboratory Animals as adopted and promulgated by the National Institutes of Health.

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